Review of the Report "An analysis in support of sediment quality thresholds for polycyclic aromatic hydrocarbons (PAHs) to protect estuarine fish"

reviewed by Gary D. Marty, D V M., Ph.D.
Diplomate, American College of Veterinary Pathologists

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GENERAL SYNOPSIS OF THE QUALITY OF THE WORK

This Report produced by the Environmental Conservation Division of NOAA sought to "use existing data on PAH effects in English sole to determine sediment PAH concentrations at which biological injury is likely to occur" [Report, p. 7]. The information presented in the Report focuses on correlative field-based studies; therefore, results from these studies cannot "adequately account for biological effects resulting from exposure to other contaminants or contaminant mixtures present in sediments" [Report, p. 8]. My critique of the Report highlights and emphasizes the limitations of applying field-based correlations for setting sediment quality thresholds.

The Report has several statements that are misleading or cannot be supported by available data.

- The Report states a cause and effect relationship between cancer and sediment PAHs, but the link between cancer and sediment PAH has not been confirmed under controlled laboratory conditions. The Report should be revised to reflect limits in the available data.

- Statements linking PAH contamination to reproductive abnormalities fail to adequately recognize the potential impact of other contaminants such as PCBs and DDTs.

- Evidence is weak for a link between adverse health effects and alterations in growth. A study of wild English sole documented increased growth in fish from a site highly contaminated with PAH; further, the only laboratory study of PAH-effects on English sole growth (peer-reviewed literature) reported no significant differences in one of two experiments.

- My 1998 analysis of English sole livers from the Hylebos Waterway provided evidence that liver-cell foci of cellular alteration (FCA) in many fish from reference sites were not diagnosed by NOAA scientists; if this underdiagnosis were corrected, and data reanalyzed, the calculated threshold for the correlation between PAH and FCA might increase.

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ABBREVIATIONS AND DEFINITIONS

The following are used throughout this Report:

DDT = a chlorinated hydrocarbon insecticide
FACs = fluorescent aromatic compounds. Biliary (gall bladder) FACs are produced by the liver during metabolism of polynuclear aromatic hydrocarbons (PAHs).
FCA = foci of cellular alteration. FCA are discrete areas of liver cells that stain different from surrounding cells. Foci are not a form of cancer, but research indicates that some types might be precursors to cancer.
PAH = polynuclear aromatic hydrocarbon.
PCB = polychlorinated biphenyls.

INTRODUCTION

The law firm Heller Ehrman White & McAuliffe requested expert review of the approach, analysis, conclusions, and relevance of the Report generated by the Environmental Conservation Division, Northwest Fisheries Science Center, National Marine Fisheries Service, National Oceanic and Atmospheric Administration, Seattle, WA, "An analysis in support of sediment quality thresholds for polycyclic aromatic hydrocarbons (PAHs) to protect estuarine fish," by Lyndal Johnson, 24 July 2000.

METHODS

For this critique, the Report and selected cited literature were examined for clarity, consistency, and appropriateness of conclusions in relation to the available data. Where relevant, alternative hypotheses are suggested. Comments listed below are roughly in numerical order; reference to the Report use a capital "R".

SPECIFIC COMMENTS ON THE REPORT

1) Background (p. 3, lines 6-8, "For example, liver cancer and related lesions have been documented in several species of wild fish *as a result of* environmental exposure to PAHs (Moore and Myers 1994) [emphasis added].") This statement implies that cause and effect for PAH exposure and liver cancer have been clearly documented , but Moore and Myers (1994) did not make this claim. Indeed, on page 364 of Moore and Myers (1994), they state that the "studies showing the strongest and most consistent ***correlations*** between exposure to particular chemical contaminant classes and prevalences of toxicopathic hepatic lesions, including neoplasms, are those on the English sole in Puget Sound [emphasis added]." Further, Moore and Myers (1994, p. 364) said that "sediment levels of PAHs *correlate* strongly with prevalences of liver lesions" and "Sediment *PCBs also correlate* with liver neoplasm prevalence [emphasis added]." Correlation is not the same as cause. This part of the Report needs to be revised to more accurately reflect available data.

2) Moore and Myers (1994, p. 366) state very definitively that "epizootiological studies of fish disease have an inherent risk of identifying chemical risk factors that are simply covariant with another unmeasured chemical. Ultimately, hypotheses based on these types of relationships can only be tested, and cause and effect established, in long-term laboratory exposures." The Report fails to provide evidence of a direct cause and effect relationship between PAHs in sediments and liver cancer. The Report reasonably addresses the potential of covariance in the middle of p. 16. Because of the potential for covariance, any conclusions can, at best, be used as a guideline; the evidence is not sufficient to conclude that PAHs alone are the cause of the observed lesions in fish sampled from the wild.

3) Background (p. 3, about 6 lines from the bottom) - "English sole from PAH-contaminated embayments ... appear to be prone to a number of other adverse health effects, including **reproductive abnormalities**, immune dysfunction, and alterations in growth and development (Arkoosh et al. 1996; Johnson et al. 1998)" [emphasis added].) The References section of the Report lists two 1998 papers by Johnson et al.; to which paper does this sentence refer? Also, the "Netherlands Journal of Sea Research" changed its name to "Journal of Sea Research" in 1997; the second Johnson et al. 1998 reference needs to be changed to reflect this change. The link between PAH contamination and "reproductive abnormalities," is not clear because other contaminants are important. For example, Johnson et al. (1998, p. 129) state that "*In statistical analyses, precocious maturation appeared to be linked to exposure to chlorinated hydrocarbons, such as PCBs and DDTs; however, additional research is needed to confirm these findings.*"

4) Background (p. 3, about 6 lines from the bottom, "English sole from PAH-contaminated embayments... appear to be prone to a number of other adverse health effects, including reproductive abnormalities, immune dysfunction, and **alterations in growth** and development (Arkoosh et al. 1996; Johnson et al. 1998 [emphasis added]"). The "alterations in growth" in this sentence are further explained by Johnson et al. (1998, pp. 130-131): "*preliminary data show that sole collected from within Eagle Harbor, where sediment concentrations of PAHs are among the highest in Puget Sound, were significantly larger at the same age than sole collected from an adjacent site outside Eagle Harbor where sediment PAH concentrations are substantially lower.*" In other words, PAH contamination is associated with **increased** growth of English sole in field studies.

5) Background (p. 3, 3rd line from the bottom) - "Liver disease, **including cancer**, is one of the most dramatic and best-documented **effects of** PAH contamination on English sole in Puget Sound [emphasis added]." As explained in my first comment (above), PAH contamination has only been **correlated with** cancer in wild English sole (Moore and Myers 1994, p. 364). This sentence in the Report needs to be revised to more accurately reflect available data.

6) Background (p. 5, last paragraph) - Experimental evidence (Rice et al. 2000) linking PAH exposure to reduced growth is equivocal. [Rice et al. was actually published in 2000, so the reference and citation in the Report need to be updated.] Rice et al. (2000) report findings from two small-scale experiments in which juvenile English sole were exposed for 10-12 days to contaminated sediments and contaminated polychaetes. One group was exposed to Eagle Harbor sediment extracts, and another group was exposed to the PAH Benzo-a-pyrene. In only one of the two experiments were growth differences significant between control and exposed fish. The paper did not report initial weights of fish in each exposure group; because only 5 or 6 fish/treatment were studied in each experiment, significant differences in initial fish size among the treatment groups could significantly affect relative growth (small fish tend to grow at a faster rate than larger fish). The Kubin

thesis has not been published in the peer reviewed literature and was not reviewed for this critique.

7) Analysis (p. 7, first paragraph) - The Report claims to focus on livers of English sole because of "the preponderance of evidence for a cause and effect relationship between PAHs and the development of liver cancer in English sole." However, the preponderance of evidence is entirely correlative, and the link between PAH exposure and cancer has not yet been experimentally reproduced under controlled laboratory conditions. As an alternative hypothesis, PAHs alone may not cause cancer, but require promotion from other contaminants such as PCBs or pesticides (reviewed by Johnson et al. 1998, p. 126). The Report needs to be revised to reflect the available data for the link between PAHs and liver cancer in English sole.

8) Analysis (p. 8, lines 3 and 4) - The report again links endpoints "such as liver cancer..." for which PAHs are known to be a strong causative factor." Based on the discussion in comment #7 above, this sentence also needs to be revised to reflect available evidence.

9) Analysis (p. 10, 1st full paragraph) - The Report points out that correlation between the low and high molecular weight analytes was too high to consider them as separate factors. What was the correlation between the PAHs and other toxicants: PCBs, chlorinated hydrocarbons, heavy metals, or other pesticides? Can these reasonably be analyzed as separate factors?

10) Analysis, DNA damage (p. 11, bottom, "A threshold in this range is also supported by a laboratory study (French et al. 1996) in which exposure to sediments contaminated with 1200 ppb dry wt PAH resulted in DNA adduct concentrations in English sole liver of **15-20** adducts/mol nucleotides...[emphasis added])." French et al. (1996) present these data in Figs. 4 and 5. Although the figures are small, the concentration of DNA adducts resulting from exposure to sediments with 1200 ng PAHs/g sediment seems to be in a range of 8-17 rather than the range of 15-20 listed in the Report. The low end of "8 to 17" reported by French et al. (1996) is considerably closer to reference DNA adduct levels (about 5) than is the range listed in the Report: "15 to 20." The Report needs to explain these apparent differences.

11) Analysis (p. 12, 1st line of 1st full paragraph) - Many authorities consider DNA adducts to represent **molecular-level** alterations rather than *tissue-level* alterations as stated in the Report.

12) Analysis (p. 12, last full paragraph) and (p. 13, middle of page) - References to Figure 3 should be changed to Figure 4.

13) Analysis, growth reduction (p. 14) - See comment # 6 above. To restate, in 1 of 2 experiments reported by Rice et al (2000), there were no significant growth differences between control and PAH-exposed English sole. Also, field experiments provide evidence

that growth of English sole was greater at a PAH-contaminated site than at reference sites (Johnson et al. 1998, pp. 130-131). Some of the findings from the Kubin (1997) thesis are described in this part of the Report, but not enough information is provided to determine if the experimental design was appropriate to support the conclusions. For example, growth can be significantly different in different tanks, and those differences can be independent of the treatment. Did Kubin's experimental design minimize experimental bias that might be a result of tank effects (e.g., were replicate tanks studied)?

14) Analysis, growth reduction, top of p. 15, "A study by Rice et al. (1999) [2000]confirmed both the effect of PAHs on growth of juvenile English sole and the importance of dietary exposure. The findings showed significantly reduced weight in juvenile English sole fed polychaete worms reared on sediments containing 3000-4000 ppb dry wt of PAHs, after an **exposure period of only 28 days**. [emphasis added]") - The experimental design described by Rice et al (2000) states that "*...polychaetes were exposed for 28 days...*" and "*Exposed worms were then fed live to juvenile English sole for 10 or 12 days.*" The Report needs to be revised to make it clear that fish in the Rice et al. (2000) experiments were fed contaminated polychaetes for 10-12 days.

15) Analysis, growth reduction (p. 15, "The percent change in weight was markedly less (0.05-0.1% per day) in exposed fish, as compared to control fish (1.1-1.2% per day).") To restate, these differences were significant only in one of two experiments reported by Rice et al. (2000).

16) Analysis, growth reduction (middle of p. 15, "The central finding from these data is that English sole exposed to sediment concentrations where toxicopathic lesions are observed are also likely to experience negative impacts on growth...") This conclusion must be balanced with actual field data summarized by Johnson et al. (1998, pp. 130-131) in which growth was greatest in English sole for a PAH-contaminated site; see comment #4 above.

17) Sources of uncertainty (pp. 15-18)- this section is well written, and the limitations of this type of analysis should be prominent in any discussion about threshold values and their application.

18) Sediment Quality Threshold Guidance (pp. 18, 2nd line from the bottom) - The report states that "the 1000 ng/g dry wt guideline does not incorporate a safety factor, as risk analyses often do, to account for uncertainty in this threshold estimate due to factors such as...." Based on the wide range of data available, including reproduction, histopathology, and chemical analysis, there are few variables that have not been accounted for. Therefore, I agree with the approach in the Report that there seems little need to include a safety factor in establishing a threshold value.

19) Effects of using inaccurate data to determine threshold values. In 1998, I examined ½ the English sole liver slides that Environmental Conservation Division of NOAA used to generate a report on fish injury in the Hylebos Waterway of Commencement Bay,

Washington (Collier, 1997). In my 1998 review, I diagnosed FCA in more than twice as many fish from the reference site Colvos Passage as did NOAA (NOAA ≈ 2.9%, GDM = 7.8%). The effect of a higher FCA prevalence in fish from the reference site is likely to increase the threshold value, but this would need to be confirmed by reanalyzing the data using more accurate histopathological data.

LITERATURE CITED

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